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INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

10/516473

Applicant's or agent's file reference PAM-010-PCT	**FOR FURTHER ACTION**	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
International application No. PCT/EP 03/05798	International filing date *(day/month/year)* 03.06.2003	Priority date *(day/month/year)* 03.06.2002
International Patent Classification (IPC) or both national classification and IPC G01N33/50		
Applicant PAMGENE B.V. et al.		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 6 sheets, including this cover sheet.

   ☒ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of 5 sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the opinion
   - II ☐ Priority
   - III ☐ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☐ Lack of unity of invention
   - V ☒ Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - VI ☐ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☐ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
24.12.2003	19.11.2004
Name and mailing address of the international preliminary examining authority: European Patent Office - P.B. 5818 Patentlaan 2 NL-2280 HV Rijswijk - Pays Bas Tel. +31 70 340 - 2040 Tx: 31 651 epo nl Fax: +31 70 340 - 3016	Authorized Officer Tuynman, A Telephone No. +31 70 340-3741

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/EP 03/05798

I. Basis of the report

1. With regard to the **elements** of the international application *(Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17))*:

Description, Pages

1-37 as originally filed

Claims, Numbers

1-32 received on 09.11.2004 with letter of 09.11.2004

Drawings, Sheets

1/10-10/10 as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

   These elements were available or furnished to this Authority in the following language: , which is:

   ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).

   ☐ the language of publication of the international application (under Rule 48.3(b)).

   ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

   ☐ contained in the international application in written form.

   ☐ filed together with the international application in computer readable form.

   ☐ furnished subsequently to this Authority in written form.

   ☐ furnished subsequently to this Authority in computer readable form.

   ☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

   ☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

   ☐ the description, pages:

   ☐ the claims, Nos.:

   ☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)).

   (Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	1-32
	No:	Claims	
Inventive step (IS)	Yes:	Claims	1-32
	No:	Claims	
Industrial applicability (IA)	Yes:	Claims	1-32
	No:	Claims	

2. Citations and explanations

   see separate sheet

Re Item V

Reasoned statement with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

Reference is made to the following documents:
D1: US-B-6 197 5751 (TANNENBAUM STEVEN ET AL) 6 March 2001 (2001-03-06)
D2: WO9902266 (VAN DAMME H.S. ET AL) 21 January 1999 (1999-01-21).
D2 -although not mentioned in the search report- was cited in the description of the present application.

The document D1 is regarded as being the closest prior art to the subject-matter of claims 1-32, and shows (the references in parentheses applying to this document):
D1 is considered to represent the closest state of the art and discloses a method for screening cellular responses of cellular components (D1, column 32, lines 29-64) comprising:
(a) providing cellular components on the surface of a solid porous substrate (e.g. filter) , said substrate having immobilised thereon an array of detector molecules (column 33, lines 34-64 or column 37, lines 14-42;
wherein
(I) said solid porous substrate retains said cellular components on its surface (column 26, lines 59-66: the pore size is so small that the cells can not pass through them and therefore they are necessarily retained on the surface of the filter) and wherein
(ii) said solid porous substrate has immobilized therein, within the pores an array of detector molecules (column 37, lines 17-21: the detector molecules are of course also present in the pores of the filter or porous material, although this is not explicitly mentioned)
(b) delivering test compounds to positions on the substrate corresponding to the arrayed detector molecules on the surface of said solid substrate (column 32, lines 32-40; example 8, column 46, lines 14-62);
(c) incubating said test compounds with said cellular components on the surface of the solid support, under conditions allowing the induction of cellular responses;
(d) assaying said cellular responses ; and
(e) identifying and characterising the cellular responses induced by said test compounds (column 33, lines 34-64 or column 37, lines 14-42.
The filter or porous substrate underneath the cells are necessarily a flow through substrate, since the whole microarray system forms a flow through system (column 9, lines 49-59). The cells are present and delivered as a culture (column 20, lines 36-63). Cell responses are assayed in this culture (e.g. example 8). Detection of the cellular

responses can take place via fluorescence measurements and/or using specific dyes (column 37, lines 14-42). In the examples mammalian cells are used. Molecules of interest can be enzymes (e.g. example 8) or other biological agents (column 8, last paragraph). The system is intended for drug screening (column 7, lines 50-53). The assaying can take place in real time (e.g. figure 8) or via end-point assaying (figure 15). Furthermore the above mentioned methods are intended for monitoring induced cellular responses of host cells (column 37, 2nd paragraph), for on-chip recombination, transformation or viral introduction of cellular components (example 9; column 4, lines 8-14) or for functional screening of cellular responses upon assaying host cells with test compounds (column 4, lines 8-14; column 7, lines 50-53). D1 also discloses a microarray for performing the above mentioned methods, wherein an array of test compounds is provided within predefined regions, said test compounds are in liquid solution and not immobilized in the substrate (e.g. column 18, line 48-column 19, line 9) or wherein a cellular component is provided on a substrate, said cellular component being conditioned for preservation on said substrate (column 20, lines 36-63), or wherein an array of detector molecules is immobilized within the substrate (column 37, lines 17-21. Furthermore the microarrays are used for providing cellular components on the surface of a substrate for use in the above mentioned methods, thereby providing said cellular components with low spreading properties (column 24, lines 54-66). Hence D1 has disclosed the elements of a kit for performing a method according to the above mentioned methods, comprising a microarray as mentioned here above.

The subject-matter of claims 1-32 differs from this known method and microarray and use thereof in that 1) the solid porous substrate is a metallo-oxide substrate with oriented through going channels and in that 2) an array of detector molecules is immobilised within the pores of the substrate.

The subject-matter of claims 1-32 is therefore new (Article 33(2) PCT).

The technical effect of this difference is that thus nutrients and other molecules can be added from underneath leading to a limited spread of the cells on the surface. Moreover, a high density array of detector molecules can be obtained, which is moreover transparent for visible light allowing for the use of optical detection techniques.
The problem to be solved by the present invention may be regarded as the provision of a microarray with low cell-spreading properties and allowing for high density of detector molecules.

The solution to this problem proposed in independent claims 1,23-28,31 and 32 of the present application is considered as involving an inventive step (Article 33(3) PCT) for the following reasons:
In order to solve the problem posed, the person skilled in the art will not be prompted by D2, which discloses a solid porous substrate being a metallo oxide substrate with oriented through going channels for the following reasons:
D2 is concerned with in vitro immunoassays which are not cell-based. There is no clue that the substrate of D2 has virtues as to properties with regard to the spreading of cells or the possibility to provide nutrients there through. Although D2 does advocate the possibility of obtaining high density arrays of detector molecules, the person skilled in the art of cell-based screening will in the first place not consider D2 since it belongs to a different technical field.

Claims 2-22 and 29,30 are dependent on claims 1 and 26-28, respectively, and as such also meet the requirements of the PCT with respect to novelty and inventive step.

The subject matter of claims 1-32 is considered to be industrially applicable in the sense of Article 33(4) PCT.

Additional Remarks

The application does not meet the requirements of Article 6 PCT, because claims 1,3,7,8,22,24,27,28 and 31 are not clear.

The meaning of "cellular components" in the above mentioned claims is not clear. To a person skilled in the art cellular components would also relate to the constituents of a cell, whereas cellular components according to the description (page 8, line 25-page 9, line 7) refer to whole cell systems. The meaning of terms in a claim should be clear from the wording of the claim alone.